WEST AFRICAN COCOA RESEARCH INSTITUTE

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WEST AFRICAN COCOA RESEARCH INSTITUTE

TAFO, GHANA

FIFTY-SECOND QUARTERLY REPORT

OCTOBER-DECEMBER, 1958

Any results recorded in this report should not be accepted as conclusive inasmuch as the report contains interim information only. More complete results are published in W.A.C.R.I. Technical Bulletins and elsewhere.

SENIOR STAFF

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Deputy Director	C.G. Johnson
Scientific Assistant to Director	G.A. Jones (returned from leave 31/12/58)
Secretary	H.L. Pugh
Accountant	H.J.S. Mill (returned from leave 14/10/58)
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TAFO STATION

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Division of Pathology	J.W. Blencowe (returned from leave 19/12/58)
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Division of Botany	D.R. Glendinning
	R.G. Hurd (returned from leave 29/10/58)
Division of Chemistry	R.H. Kenten
	R.K. Cunningham (returned from leave 8/10/58)
Division of Agronomy	R.W. Smith
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IBADAN SUB-STATION

Deputy Director (Nigeria)	T.W. Tinsley
Entomologists	R.G. Donald
	C.K.H. Martini
Pathologists	J.M. Thresh
	D. Kay (returned from leave 21/11/58)
Agronomist	J.F. Longworth

WEST AFRICAN FUNGICIDES RESEARCH UNIT

Mycologist	E.C. Hislop (on leave 17/12/58)
Chemist	P.O. Park.

STAFF MATTERS

Dr. J.W. Blencowe, Mr. G.A. Jones, Mr. R.K. Cunningham, Dr. R.G. Hurd and Mr. D. Kay (Nigeria) returned from leave during the quarter. Mr. E.C. Hislop, the mycologist of the West African Fungicides Research Unit at Ibadan, went on leave in December, 1958.

VISITORS

Mr. V. Mukete, Minister of Research and Information for the Federation of Nigeria visited the Institute in November. Other visitors included Mr. K. Twum-Barima, Head of the School of Agriculture, Kumasi College of Technology; Mr. & Mrs. Zvi Halmer of the Israeli Embassy, Accra; Mr. P. Terver, Director-in-charge of African Affairs, F.A.O.; Mr. K. Hackman of the London Office of the Ghana Cocoa Marketing Board; Mr. Clarke, U.K. Ambassador to Liberia; and members of the Netherlands and Italian Missions to Ghana.

Dr. E.J. Hewitt of Long Ashton Research Station spent several weeks at the Institute to study the work being done on cocoa nutrition, and to advise on the future programme.

Visitors to the Sub-Station have included Mr. du Plessis of the Division of Plant Control and Quarantine, Pretoria; Lord Milverton; Mr. M. van Bottenburg, Tropical Institute, Amsterdam; and Mr. T.B. Oluyide, Nigerian Produce Marketing Co. Ltd.

MANAGEMENT AND MAINTENANCE

Construction work on the Spectrographic Laboratory has been virtually completed, and the equipment has been installed.

At the Ibadan Sub-Station one Senior Staff house has been completed, and the second is nearing completion. Six new insectaries have been erected.

CROP DELIVERY

The number of pods harvested during the quarter is given below:

Month	Old Station	Square Mile	Total
October	53,102	119,239	172,341
November	127,009	210,737	337,746
December	72,507	115,494	188,001
Total	252,618	445,470	698,088

SWOLLEN SHOOT DISEASE

The number of trees removed during the quarter due to swollen shoot and other causes is given below:

Month	Trees destroyed		Total	Total Seedlings removed
	Swollen Shoot	Undiagnosed		
October	32	-	32	32
November	14	2	16	33
December	38	10	48	19
Total	84	12	96	84

UPPER AMAZON SEED

A total of 52,745 F3 Upper Amazon pods were sent to Nigeria during the quarter.

METEOROLOGY

Month	1958		20-year mean	
	Rainfall (in.)	Wet days	Rainfall (in.)	Wet days
October	6.49	20	8.90	18.7
November	3.12	17	4.12	10.9
December	3.32	7	2.38	6.2
Total	12.93	44	15.40	35.8

T A F O S T A T I O N

PATHOLOGY DIVISION

VIRUS RESEARCH

A further visit was paid during the quarter to the outbreak of cocoa virus disease discovered at Gandorhun in Sierra Leone early this year (Quarterly Report No. 50, p.4). The number of infected trees had increased by some 50 per cent, but the symptoms of infection had become less easy to see, and there was less evidence of reduction in vigour of the infected trees than there had been six months earlier.

The routine developed for the estimation of the effects of virus infection on growth-rate of Amelonado cacao seedlings has proved to be of practical value, as well as yielding information of academic interest. The Gandorhun isolate mentioned above has been compared with severe and mild virus of the New Juaben type and the results confirm the observations in the field. The reduction in leaf area, fresh weight of leaves, height and total fresh weight of seedlings infected with these isolates when compared with control were recorded 12 weeks after infection and are tabulated below.

Isolate	Percentage reduction relative to healthy controls			
	Leaf area	Fresh wt. leaves	Total fresh wt.	Height
Gandorhun	18.9	32.7	28.7	13.1
Mild 1A	15.4	22.3	21.3	8.4
Severe 1A	36.8	49.7	41.9	29.7

The Gandorhun isolate is intermediate between the mild and severe New Juaben isolates in its effect on the growth of seedlings and it may be considered a relatively mild virus.

The effects of the unrelated virus isolates from Kpeve, New Juaben and Mampong, when present singly and in combination, on the growth of Amelonado seedlings have now been determined (cf. Quarterly Report No. 51). In these experiments test plants were infected at the bean stage and the growth attributes of treated plants were recorded at two-weekly intervals. The reduction in leaf area, leaf fresh weight, total fresh weight and height recorded 16 weeks after infection and expressed as a

percentage of the healthy controls are tabulated below.

Isolate	Percentage reduction relative to healthy controls			
	Leaf area	Fresh wt. leaves	Total fresh wt.	Height
New Juaben	38.2	37.4	42.4	32.7
Kpeve	56.5	53.8	26.8	34.0
Mampong	18.9	10.2	22.4	29.4
New Juaben and Kpeve	63.1	64.2	40.5	44.9
New Juaben and Mampong	63.9	62.0	49.5	40.8
Kpeve and Mampong	61.7	59.9	35.3	29.1
New Juaben, Mampong and Kpeve	64.1	64.0	45.4	37.3

Mutual protection has not been demonstrated between the Kpeve, New Juaben and Mampong isolates, and when present in combination produce cumulative effects on the growth of Amelonado seedlings (cf. Quarterly Report No. 51, p.19). The effect of other related and unrelated virus isolates when present in complex on the growth of cacao seedlings is now being investigated.

Insect transmission studies

Experiments to determine the extent of differential transmission of 14 cacao virus isolates by 10 vector species have continued (Quarterly Report No. 51).

The Yakasse and Gandorhun isolates have been transmitted by F. virgata (Ckll.), the Nkawkaw, Bisa, Alaparun, Offa Igbo, Amafie, Gandorhun and Yakasse isolates by Planococcus kenyae (Le P.) and the Yakasse and Amafie isolates by P. convacocerarii James.

Results are incomplete, but those to date indicate that on the basis of differential vector transmission three groups of isolates may be readily distinguished. The first group contains isolates which are transmissible by F. virgata (Ckll.), but not by P. adonidum (L.). These isolates characteristically induce swellings and usually leaf symptoms on infected Amelonado cacao. The second group contains isolates which have been transmitted by P. adonidum, but not by F. virgata. Isolates included in this group characteristically induce leaf symptoms only, although it has been repeatedly confirmed that the Mampong isolate is also referable to this group. An isolate

of the third group, that from Asalu in Nigeria, has not been transmitted by any of the 10 mealybug species. No differences have been domonstrated between isolates of the swollen-shoot type by any of the 10 vector species. Similarly, no differences have been shown to exist between isolates of the mottle-leaf type. Differences in vector transmissibility are of use only in distinguishing between these three groups and are of no use in distinguishing between isolates within a group. The classification of cacao virus isolates into three viruses (Quarterly Report No. 47 & 50) is therefore supported by these vector differences although the status of the Mampong isolate which shows similarities to both isolates of the cacao swollen-shoot virus and the cacao mottle-leaf virus, remains obscure. These results indicate also that vector specificity is so far confined to the vectors F. virgata and P. adonidum.

The severe New Juaben virus isolate has been transmitted occasionally by Formicoccus tafoensis. The ability of this species to transmit a cacao virus isolate has not hitherto been reported.

Resistance and tolerance

The testing of cocoa varieties for resistance and tolerance to infection with cocoa swollen shoot virus continues, using seedlings infected at the bean stage. Seedlings growing in baskets are now being prepared for planting in the field in 1959; these will be used for the complementary experiments on the resistance and tolerance of these varieties under field conditions. It is hoped that about five acres will be planted during 1959 and 1960.

STUDIES ON BLACK-POD DISEASE

Fungicide spray control trials

Numbers of diseased pods on the three fungicide experiments (cf. Quarterly Report No. 49, p.5) have not exceeded 4 per cent of the total pods harvested during 1958 under any treatment including the unsprayed controls. This has occurred despite the restriction of harvesting to six-weekly intervals. The primary objects being to study the effects of sprays applied at different times of the year and the effect of copper on total yields, both the main trials will be continued in 1959. Four years' data will then be available (two years pre-treatment and two years under treatment) which should be sufficient to show valid treatment effects with individual plots one-quarter acre in size. It has not yet been decided whether to continue the third experiment, which includes the fertilizer treatment.

Pod susceptibility experiment

Incidence of wilt on the sprayed trees during 1958 (cf. Quarterly Report No. 51, p.6) has been much higher than expected, 62 per cent of all fruits set compared to 53 per cent on the unsprayed trees. In the previous two years no difference was recorded in percentage wilt for sprayed and unsprayed trees. The difference now noted is not entirely accounted for by the reduction of Phytophthora infections on cherelles on the sprayed trees. The copper spray has definitely increased fruit fall or cherelle wilt, although the number of fruits set is not affected. The crop during 1958 was again borne on a completely different set of cushions compared with the previous two years.

Resistance and tolerance

Tests on tolerance measured by the rate of growth of pod lesions following standard artificial inoculations, have shown marked differences between selections included in the 1st Clonal Trial. If the selections are ranked for tolerance the selections at each end of the table are those which are apparently also resistant or highly susceptible, respectively. It will, however, be some time before the analysis of field data for resistance is completed and before it is known how closely the rankings for tolerance fit those for resistance.

Strains of Phytophthora palmivora

Investigation into the possible occurrence of different strains of P. palmivora in Ghana is being continued (Quarterly Report No. 46, p.6). Isolates from 28 widely separated localities are being tested by natural and artificial culture techniques. Comparison of the Ghanaian fungus with those of some other West African territories is also being made. (Rep. W. Afr. Cocoa Res. Inst. 1956-57, p.36).

Saprophytic survival of P. palmivora

Work is in progress to determine the means by which P. palmivora survives between crops. Preliminary investigations indicate that some fungistatic property exists in the soil, and the nature of this property and its relation to survival of the fungus in soil are being investigated. Soil, bark, detritus from branch junctions, and foliage have also been sampled for the fungus at regular intervals, and sampling is being continued.

MYCORRHIZA

Methods of direct isolation, without use of surface sterilizing agents, of a mycorrhizal fungus from roots have so

far been unsuccessful. Complete control of the rhizosphere bacterial population was obtained with aureomycin. Fungi isolated from root surfaces include Aspergillus niger, Clodosporium herbarum, Trichoderma viride, and species of Cephalosporium, Chalaropsis, Cunninghamella, Fusarium, Hormiscium, Penicillium, Pestalotiopsis, Spicaria and Rhizoctonia.

ENTOMOLOGY DIVISION

MEALYBUG STUDIES

Oyoko

Following the second application of the double dieldrin treatment (Quarterly Report No. 51, p.8) crematogasterine ant populations fell to a lower level than that recorded after the initial double treatment with dieldrin. This is probably because the nests individually had not grown to pre-treatment size though they had regained pre-treatment numbers, and the closer spacing of the two applications in the second treatment may also have been more effective.

Except in three small farms lying close together on the margin of the treated area, populations of Pseudococcus njalensis Laing fell to zero (as recorded in counts to hand-height) during the second month after completion of the dieldrin treatment. Counts on complete trees (felled as virus contacts) followed a similar pattern, P. njalensis populations declining to a very low level. In the last two months of the quarter only three trees were recorded with more than one adult P. njalensis.

The high counts made on the three marginal farms are perhaps the result of incomplete spraying; during the second treatment certain small food-farms enclosed in the treatment area could not be sprayed due to the damage which would have been caused to growing crops (maize and cassava).

Ant/Mealybug Control Trials - Series II

No differential effect has been detected between the dieldrin and dieldrin/coumarone treatments in the ant data. The reasons for this are being investigated in a small field trial designed to compare the efficiency of different dieldrin/coumarone formulations and methods of application.

A preliminary experiment using ant-established colonies of P. njalensis on seedlings has been set up to test the efficacy of the following systemics against mealybugs: Rogor 40, Ekatin

and Ciba 570. Soil, stem and foliage treatments are being studied separately. It is hoped that these studies will later be extended to the partial systemics - Diazinon, Folidol and Diptrex.

Pod fumigation

Pods sent out of Ghana for use as seed are at present dipped in a solution of mercuric chloride (approx. 0.4%) to kill fungus spores. It has been found that this treatment is not adequate to kill mealybugs harbouring in the pods, particularly those which colonise Characoma mines. Attempts have therefore been made to develop a method for fumigating pods, with the following results.

Cocoa sacks, approximately 5 cu. ft. in volume, were provided with a polythene lining and each filled with approximately 170 pods. In each sack three pods infested with mealybugs (placed beforehand in artificial mines made by a cork-borer) were placed in the bottom, middle and top sections of the sack. The sacks were then tied and sealed after a wad of mutton cloth saturated with fumigant had been dropped into the top of the sack.

The fumigant used was a 50/50 mixture of ethylene dichloride and carbon tetrachloride with a range of 1 oz. to 9 oz. per sack. The pods and fumigant were left for 48 hours in the sealed sacks, stacked in an open shed.

The results were somewhat variable owing to tears developing in the polythene liners, but briefly, it may be said that 100 per cent kill was obtained with the 5-7 oz. treatments, as against a 26 per cent mortality in the untreated controls. Germination tests showed no ill-effects from fumigation at the 5 and 6 oz. per sack levels.

The method is very promising, but improved liners are necessary.

MARMARA STUDIES

Identifications of the two major parasite species of Marmara (Lepidoptera, Gracilariidae) have now been received from the Commonwealth Institute of Entomology. They are;-

Bracon sp.	(Hymenoptera, Braconidae)
Ageniaspis sp.	(Hymenoptera, Encyrtidae)

The latter species, which is responsible for the bulk of Marmara parasitism at Tafo, belongs to a genus the members of which are probably wholly polyembryonic. The few observations made on the

present species indicate that it conforms with the general habits recorded for this genus: several parasite larvae are to be found in each host larva (up to 24 have been observed in one host) and the parasitised final stage Marmara larva is larger than its healthy counterpart. Parasite cocoon formation has been observed only in final instar host larvae.

MINOR PESTS

Cerambycidae

Work continues on the biology and taxonomy of the Tragocephala species associated with cacao in West Africa. It is apparent that there is some taxonomic confusion and the results of this aspect of the investigations will be reported in due course.

A simple technique for the laboratory rearing of Tragocephala has been established. Adult insects, enclosed in lamp-glass cages, are fed on pieces of fresh green chupon. Mating occurs freely within the cages, oviposition taking place on the food material. Short lengths of chupon tissue containing eggs are retained in closed petri dishes till eclosion of the 1st instar larvae. These, on emergence, are placed in artificial burrows made in partially dried green wood, wilted and less sappy tissue being necessary for the survival of young larvae. Subsequently they are transferred to thicker lengths of slightly older fresh wood, this being renewed as soon as a larva has burrowed down its length. On pupation they are removed to hollowed stems of dry wood to await emergence of the perfect insect.

Leaf-eating Lepidoptera

Approximately 75 species of lepidoptera have been recorded as being associated with cacao in West Africa, but at W.A.C.R.I. alone there are approximately 100 different specimens of leaf-eating lepidoptera. The rearing of as many of these species as possible is in progress and the adults are sent to the British Museum for identification.

AGRONOMY DIVISION

On the Uniformity Trial Area, Old Station, harvesting of the main crop has continued. Over the quarter the percentage of discarded pods was 3.7, and the total yield of dry cocoa to date is nearly 15 per cent higher than during the corresponding period of the previous year.

Investigations have been started on soil moisture as a limiting factor to growth during the dry season. A pilot irrigation trial has been set up, in which cocoa grown at two soil moisture levels, both within the wilting point - field capacity range and maintained by irrigation, is compared with non-irrigated cocoa. Amazon hybrid varieties planted out in 1958 are being used, with 30 trees per plot. Similar investigations are also being carried out using potted seedlings.

BOTANY DIVISION

PLANT BREEDING AND SELECTION

In the previous Quarterly Report it was mentioned that favourable reports had been received from British and American panels on the quality of some of our new cocoa varieties. A corresponding report has now been received from the Swiss manufacturers, which is in fair agreement with that of the British panel. In these two cases the cocoa was assessed for correspondence with normal West African Amelonado, but the American manufacturers have assessed for quality, within the context of the West African group of cocoas, and their results are therefore somewhat different from those of the other panels. In particular one variety which was regarded as distinct from normal West African cocoa by the C.S.A.C. and the Swiss manufacturers and as inferior by the latter, received a favourable report from the Americans.

The British and Swiss manufacturers each submitted a joint report of their constituents, but in the American report the opinions of individual firms were recorded. There appeared to be a wide divergence of opinion among these firms, and a statistical analysis (by Mr. de Jonge) showed no significant concordance between the rankings given the samples by the various firms. When the opinion of one manufacturer was omitted a measure of concordance was found but a considerable divergence of opinion remained even within the reduced group. It seemed possible to account for this as a variation in preferences for two characters, one of which is "flavour", to which different manufacturers appear to attach varying degrees of emphasis, and the other is as yet unidentified. The "flavour" character in this context refers to the presence of a flavour additional to that of normal West African cocoa.

Records of stem height, stem diameter, and number of leaves were taken on the F1-F2-F3 trial (see Quarterly Report No. 51) in May, i.e. shortly after planting, in August and in November, 1958. The seed was planted between 30th November and 18th December, 1957. As the position of the seedlings in the nursery may have affected their size at planting, comparisons of the different generations in August and November have been based on data corrected for the figures in May by covariance analysis. No clear evidence of differences was found in August, but in November small but statistically very highly significant differences were apparent, with the F1 generation growing most vigorously and the F3 least vigorously.

In an F2-F3 trial at Bunso, planted in 1953, the total number of pods harvested from the F2 generation exceeded that from the F3 generation, up to the end of the 1957-58 season. By the end of November, 1958, however, the F3 generation of types T60 and T63 had caught up with the F2 in total yield, though the F3 generation is still lagging in the other three types included in the trial.

An examination of the growth measurements on the 1957 - planted Series II trials in Ghana has indicated significant differences in the placings of the varieties relative to each other in the Fumso and Foso trials. These differences may be due to varietal preferences for one site or the other, but they could alternatively be due merely to unequal treatment of the varieties as seedlings in the nursery before planting out. A further set of measurements will allow such spurious effects to be eliminated by covariance analysis. In the Buaku trial there is some indication that the age of the seedling at planting may affect the subsequent growth rate.

PHYSIOLOGY OF NUTRITION

A series of nutrient culture experiments are being set up, in accordance with the recommendations made by Dr. Hewitt of the Nutrition Unit, Long Ashton Research Station, during his visit to the Institute. In particular, an investigation of the effects of boron and copper deficiencies on cocoa growth is being initiated. Cocoa seedlings grown under shade in concrete pots at W.A.C.R.I. have developed malformations, which resemble the symptoms of "stag's head" and "paint-brush" disease, reported principally in Nigeria (e.g. R. Bull, "A report on a new disease of cocoa which has appeared in the Ibadan area of the Western Region of Nigeria", 1955). It is considered that these symptoms

may be due to mineral deficiencies, and an attempt is therefore being made to reproduce them in culture solutions.

The potted seedlings showing these symptoms at W.A.C.R.I. have been treated with solutions of either boron, copper or zinc, and with solutions of the three micronutrients together. It is, however, difficult to say whether these treatments have been effective, as the symptoms have considerably reduced with the onset of the dry season. This temporary disappearance of symptoms seems also to be a feature of "paintbrush" and "stag's head" disease.

In the nutrient culture experiments an attempt will be made to induce boron, copper and boron + copper deficiencies. The seedlings will be divided into three sets, one of which will be grown in the deficient culture solution from germination, while the other two sets will be grown in a complete nutrient solution for two and six months respectively, before transfer to the deficient solution.

Both sand and water culture will be used, and the experiment will provide an opportunity to compare these two methods in respect of the general vigour of the seedlings, and the ease with which deficiency symptoms can be induced.

The local white sand which is being used has been chemically cleaned by a method developed by Mr. de Endredy of the Soil and Land-Use Survey Department of the Division of Agriculture. A sample of the treated sand is being analysed by the Department of Geological Survey. Careful precautions, such as the removal of the copper insect-proofing from the insectary where the cultures are to be set up, and the use of polythene for solution storage, are being taken to avoid contamination.

CHEMISTRY DIVISION

SOIL FERTILITY

Field Experiments

Harvesting of the field experiments is not yet complete, but preliminary reports suggest that tendencies will be similar to those of last year. The young Amazon cocoa established on clear-felled land at W.A.C.R.I., and treated with mulch, has produced to date, in its second year in the field, 150 lb. dry cocoa per acre. On very badly degraded soil a Guatamala grass mulch together with a NPKMg fertilizer mixture

has brought young Amazon cocoa into bearing after 18 months in the field. On the Shade and Manurial Trial even larger yields than those obtained during the 1957-58 season are being harvested.

Pot experiments

Iron deficiency has been produced in young cocoa seedlings by liming a normal cocoa soil. This can only be remedied by spraying the leaves with 1 per cent ferrous sulphate solution.

COCOA PREPARATION AND QUALITY

Work on methods of fermenting and drying small quantities of beans is continuing. During the main crop particular attention has been given to the effect on the quality of the finished cocoa of fermenting small heaps (less than 30 lb.) under conditions where the external temperature is held closely to a constant value.

Experiments have also been made to find out how far the heat produced when cocoa beans are heaped is dependent on microbial activity in the pulp.

I B A D A N S U B - S T A T I O N

PATHOLOGY SECTION

VIRUS RESEARCH

Observations on swollen shoot outbreaks in the abandoned areas of mass infection

For some years all the trees in several swollen shoot outbreaks in the Ibadan and Ilaro abandoned areas of mass infection have been inspected for virus symptoms at monthly intervals. The information obtained is summarised under the following sub-headings.

Symptom expression:

Saplings infected with typical isolates of cacao swollen shoot virus first show transient red vein banding symptoms on the flush leaves, or speckled clearings and angular chlorotic flecks along the minor veins of hardened leaves. Both transient and permanent leaf symptoms intensify on succeeding flushes, but then become restricted to the principal veins as the plants pass into the chronic stage of infection and swellings appear. The sequence of symptom expression on naturally infected mature trees is often similar, although the acute and chronic phases are less distinct. Moreover, it is often difficult to see symptoms due to the poor condition of the trees, their limited growth at certain seasons and the height of their canopy branches. Furthermore, the trees are

not all uniform in their reaction to infection and different virus strains may occur in trees within quite restricted areas. Thus it is hardly surprising that there were differences between the symptoms found on infected trees in the field, at the time they were discovered and subsequently.

At each observation plot leaf symptoms were usually the only ones found on trees discovered at an early stage of infection. Symptoms were then often restricted to fan branches produced at periods of vigorous growth. At other times symptoms were found only on the few chupons which were flushing, usually from the base of the tree or the main branches. Leaf symptoms usually became more extensive on subsequent inspections and swellings often appeared. However the leaf symptoms on some trees were missed until they had already become extensive and in some instances accompanied by swellings. These appeared, on most of the trees at Offa Igbo, an average of 3.4 months after leaf symptoms had first been recorded. Swellings were also common at Koroboto, but rare on the trees at Ajia, where they did not appear until an average of 11.3 months after mosaic symptoms.

The differences between the plots were partly due to the efficiency with which the trees could be examined for symptoms. At Offa Igbo, the canopy was broken by capsid damage and die-back and most of the living branches could be seen readily, whereas the trees at Ajia had high inter-locking canopy branches and swellings were particularly difficult to see from ground level. Furthermore, transmission tests showed that typical virus isolates from Offa Igbo caused more conspicuous and numerous swellings than ones from Ajia. At both plots and at Koroboto some isolates caused severe leaf symptoms and swellings on test seedlings, whereas others caused only swellings or mosaic. The mosaic symptoms were sometimes very mild and at Koroboto 10 per cent of the infected trees seemed healthy at each inspection and only showed inconspicuous chlorotic specks on rare occasions. They were then restricted to the leaves of only a few branches. Even the most conspicuous infections sometimes became almost symptomless, particularly as leaves did not persist long under the relatively open conditions at most of the plots. Furthermore, branches with symptoms were often lost during drought periods or when capsid damage and die-back were particularly severe.

The rate of virus spread:

The presence of virus may be demonstrated by transmission tests carried out with mealybugs or grafts, which are difficult on a field scale and unreliable, particularly with symptomless infections. Chemical methods of diagnosis are similarly unsatisfactory and coppicing is the only available method for the large-scale indexing of trees, although it suffers from the disadvantage that it checks spread. Consequently, natural spread can only be followed by observations on the appearance of trees with their first symptoms, an unsatisfactory method further complicated by the variable latent period of infection, ranging from a few weeks to many months and influenced by seasonal factors, the growth and condition of the trees and the ease of symptom detection. Nevertheless, certain trends are common to observations in Ghana and Trinidad.

At each plot the number of infected trees increased annually and followed a sigmoid trend with time. Thus at Koroboto and elsewhere the annual number of apparently new infections increased until approximately half the trees were diseased and then decreased, as infection spread to the edge of the plots. A more realistic estimate of spread was obtained for a longer period when percentage infection was expressed in multiple infection units.

The annual number of new infections is not proportional to the number of existing ones and the ratio between the two decreases with time, suggesting that on this criterion rate of spread decreases as outbreaks increase in size. This is to be expected from the available information on the movement of the mealybug vectors, which is usually for limited distances between the canopy branches of adjacent trees. Thus mealybugs are unlikely to reach healthy trees from an increasing proportion of the infected ones in an outbreak and most spread occurs from those on the perimeter; particularly as they are likely to be in an early stage of infection and support higher mealybug populations than chronically infected trees.

Rate of virus spread is difficult to compare between plots because of differences in spacing, size and condition. However, there were considerable differences in the time taken for outbreaks to show similar increases. Thus the number of infected trees at Offa Igbo increased from 50 to 100 in four months and from 50 to 200 in sixteen. Comparable increases occurred at Koroboto in thirteen and twenty-two months, respectively. Virus spread even less rapidly at Iweke where the number of infected

trees in a two-acre plot only increased from 13 to 84 in six years. Even trees in contact with infected ones may not show symptoms for some years. This situation should be compared with the spread of aphid-borne virus diseases of certain annual crops such as sugar beet, groundnuts or brassicas which may be almost completely infected in a few months.

At each plot the number of known infections did not increase regularly and more infected trees were discovered at certain times than at others. This could indicate peak periods of spread some months previously. However, a more likely explanation is that symptoms were most extensive and conspicuous when the trees were growing vigorously. Thus relatively large numbers of trees were discovered at the main flushing periods in March-April and October-November, at the beginning and end of the rains. By comparison relatively few infections were discovered in the cool cloudy conditions of July, August and September, when trees in West Africa make little new growth.

The pattern of virus spread:

The pattern of virus spread at each of the Nigerian observation plots was similar to that described in Ghana and Trinidad and most new infections were found alongside existing ones. However, spread was not uniform in all directions from the original source and the outbreaks were irregularly shaped. Furthermore, outlying infections appeared; presumably caused by wind-borne mealybugs. Thus at Koroboto 57 per cent of the 135 infected trees found in 1954 and 1955 were within five yards of obviously infected trees and 88 per cent were within twenty. The corresponding figures for 1956-57 were even higher (84 and 100 per cent) respectively as relatively few susceptible trees occurred outside twenty yards. The extent of spread between adjacent trees was probably even greater than is apparent from these data as some sources of infection were symptomless.

The limited spread and steep gradients of infection around swollen shoot outbreaks are consistent with the results of coppicing experiments on the distribution of new infections around other outbreaks in Nigeria. Moreover, they emphasise the importance of local spread by mealybugs moving through the canopy, and give a further indication that the spread of swollen shoot disease into new plantings and plots of mature cacao can be checked without difficulty, if they are not contiguous with infected trees and inspected and rogued at regular intervals.

Cacao Necrosis Virus

On Amazon seedlings grafted five months ago with patches from Cola nitida permanent leaf symptoms have appeared on one leaf but not on subsequent leaves. It would still seem premature to consider the experiments on the presence of CNV in Cola nitida as conclusive.

The available data on spread of CNV in the field has been checked critically and it is now clear that the recording has been too inaccurate to determine whether the virus spreads slowly or not at all. The only known outbreak of the virus will be re-numbered and new observations started as soon as possible.

Tests to transmit CNV with various species of insects have been resumed, as it will now be possible to observe the test plants over longer period.

Graft transmission of cacao viruses using leaf tissue

Experiments to transmit cacao viruses by inserting pieces of leaf tissue under bark flaps were started with CNV and have now been done with the CSSV isolate from Balogun. The method was first developed by Wallace (Phytopathology, 37, 1947) in studies on the transmission of citrus psorosis virus. These results have been confirmed using small lime seedlings and two Nigerian virus isolates from citrus. Cacao seedlings about 12 months old were grafted with leaf tissue from seedlings infected with the Balogun virus isolate. Seedlings that were cut back immediately after grafting produced symptoms 6-8 weeks later whereas seedlings that were left intact were still symptomless 18 weeks after grafting. Seedlings four weeks old produced symptoms after 8 weeks and if defoliated, they died within 4 weeks. Two-week-old seedlings showed symptoms after 6 weeks. If pieces of leaf tissue were inserted into the radicles of beans, leaf symptoms appeared during the 5th week after inoculation.

The percentage number of infected plants in experiments using young plants of 4 weeks old and less, was rather low. This may have been as a result of lack of skill but it seems also possible that young plants are more difficult to infect by grafting than by infective insects.

Preliminary observations indicate that pieces of fully hardened leaves showing permanent symptoms are a more suitable inoculum than pieces from immature leaves showing the red vein banding symptom. Experiments are in progress to test whether the leaf tissue grafting technique can be used to assess the presence of transmissible virus in various parts of infected plants.

Transmission of cacao virus by parasitic plants

Experiments on similar lines to those reported from Tafo in Quarterly Report No. 51 have been carried out in Ibadan. The parasites Cassytha filiformis (Lauraceae) and Cuscuta subinclusa (Convolvulaceae) have been used on cacao plants infected with Abuku, Ikire, Alaparun, Olanla, Koroboto and Asijere isolates. No transmissions were recorded when healthy plants were subsequently parasitised.

STUDIES ON "DIE-BACK" OF CACAO

The effect of fungicidal sprays and pruning on "die-back" in cacao

In a plot of over two hundred mature cacao trees, the following treatments have been compared for a period of ten months:

1. Pruning away all existing areas of "die-back" each month.
2. Pruning combined with monthly high-volume spraying of a 3% solution of a mercury-copper fungicide.
3. Pruning combined with a monthly high-volume spraying with a 6% solution of a copper fungicide.
4. No pruning and no fungicidal spraying.

The trees were all sprayed to control capsids. The number of new areas of "die-back" formed was recorded monthly for each of the treatments. At the first examination an average of nine new "die-back" areas per tree was found in all the treatments. This number decreased at each of the four following examinations and afterwards remained between the average of 1.4 to 2 areas per tree. There was considerable tree to tree variation, but there was a tendency for "die-back" to be less on the pruned and sprayed trees. Cultures of the fungi occurring in the "die-back" made after one, four and ten months indicated that there was no consistent reduction of Calonectria rigidiuscula in the sprayed plots. Regular spraying against capsids so reduces the number of new "die-back" areas formed that it masks the effects of other treatments.

A considerable measure of control of black pod was found on the sprayed trees, but no difference was found between the mercury-copper and the unsynergised copper spray.

The effect of irrigation on "die-back" of mature cacao trees

Thirty-seven trees in a plot of seventy-four trees have been irrigated for one year. Water has been applied by hose at

five day intervals, each application being equivalent to one inch of rain. Monthly records were made of the number of new areas of "die-back". The trees were sprayed to control capsids. It is significant that the effect of irrigation on "die-back" was not masked by capsid spraying. "Die-back" occurred consistently on the irrigated trees, and the greatest difference, relative to the non-irrigated trees, appeared during the drier months of February, March, April, August, September, October and November.

ENTOMOLOGY SECTION

SHOT-HOLE BORERS

Two experiments have been laid down to investigate the incidence and some possible methods of control of shot-hole borers (Scolytidae) in seedling cacao, and the effects of borer attack on the plants. The insecticidal preparations used are a urea-formaldehyde lacquer containing 10% dieldrin and 4% aldrin when dry, and dieldrin emulsion at 0.5%. In preliminary tests of phytotoxicity, the lacquer formulation was found to be unsuitable for application to unhardened tissues, which were killed within four days of application; no apparent damage was done to hardened stems and seedlings could be treated on the lower portions of the stem.

In one experiment at Ibadan the effects of a single treatment of the lower portions of the stem with the lacquer formulation is being compared with the use of an aqueous emulsion of dieldrin applied only to the lower 12 inches at monthly intervals. Control plants receive no insecticide. The seedlings are not being watered during the dry season, though they were liberally watered during the first fortnight after planting in October.

Shot-hole borer damage was first noted in mid-November and by the beginning of December 15.4% of the 480 seedlings in the plot had been attacked. This attack had increased to 34.8% of all seedlings by the end of the month, being slightly heavier on those sprayed with emulsion than on the untreated ones (41.8% and 36.7% respectively) but some control was obtained using the lacquer (24.2% attack).

Similar results have been obtained at Akure where a larger trial was laid down using 1,200 seedlings. Here a third

insecticide treatment of 0.5% dieldrin emulsion has been included and applied to the whole plant as opposed to the lower portions of the stem only. The percentage of seedlings attacked was as follows:

lacquer: 13.7%
aqueous emulsion up to 12 inches: 30%
aqueous emulsion overall: 21.7%
untreated 26.3%

In this experiment half of the plots in each treatment are receiving 1 gallon of water per plant twice weekly during the dry season. As the rains continued well into December at Akure watering was not started until the middle of that month and no effect has yet been noticeable.

AGRONOMY SECTION

Agodi Cacao Experimental Farm

The seedling nursery has been extended, and pods of supposed virus-tolerant Amazon varieties have been planted in preparation for a field experiment later in the year. Some early losses were experienced, but the seedlings are now growing satisfactorily.

A nursery trial has been laid down with the object of selecting a method of producing uniform, vigorous seedlings for planting in future field experiments. Present nursery practice in Nigeria is to grow cacao seedlings in whalehide pots closely packed in the beds, the nurseries being heavily shaded. Seedlings produced by this method are often very variable, and establishment losses from this and other factors are high. The experiment consists of eighteen replications of a 2^2 factorial layout with NPKMg fertilizer mixture application versus none, and close packing of pots versus wider spacing. Six replications have been placed under each of three levels of shade; no shade, light shade and heavy shade. Fifty seedlings have been used per treatment, from unselected F3 Amazon seeds planted in a brown, friable loamy soil in whalehide pots. These seedlings will be planted in the field in May, in the form of a randomised block design, with six replications of the twelve treatment combinations. The effects of the various treatments on growth of the seedlings in the nursery will be measured before planting in the field, and their effects on establishment in the field will be

observed.

Koroboto

In general, growth in the two seedling experiments has been satisfactory, although symptoms of mineral deficiency, including abnormal leaf shades have been noted. These have not been confined to the non-fertilized plots. A proprietary fertilizer mixture "Sequestrene" was applied to the fertilized plots in an attempt to correct the supposed minor element deficiencies, and in most cases this has been successful. Another feature common to both experiments was that in a large number of seedlings the apical bud failed to produce the normal flush of leaves. The young leaves died, either in the bud, or soon after they appeared, and the stipules remained attached to the stem, the result being the typical "paint-brush" condition. Later, an axillary buds became dominant and grew on in the normal way. This prevented the seedlings producing a jorquette at the usual stage, and they are now abnormally tall. Spider mite and Earias biplaga Wlk. have been common pests, the former appearing on less vigorous seedlings, and causing considerable damage.

Plants infected with mild and with virulent virus are now producing leaf symptoms, and in a few cases, swellings. Calonectria inoculations have been made using a modification of Owen's technique. A puncture was made in the stem with a hot needle before inserting a needle which had been dipped in a suspension of Calonectria spores and mycelium.

Two months after planting it was found necessary to provide shade over the first experiment, as the seedlings were losing their leaves rapidly with the onset of drier conditions. A palm-frond canopy was erected. Similarly a light canopy was erected over the "no shade" plots in the second experiment, and the canopy over the shaded plots has been increased.

WEST AFRICAN FUNGICIDES RESEARCH UNIT

The combined assay technique, has been in use continuously during the period under report. Copper fungicide spray deposits on cacao pods are determined chemically and, simultaneously, the prophylactic efficiency against an artificial infection with black-pod disease is measured. Various small modifications to both the chemical and bioassays have been introduced to produce more consistent results.

The whole technique can be summarised by the following time-table:

Day 1: Collection of nearly mature green pods, taking care to avoid bruising. Application of fungicide, using the laboratory sprayer.

Day 2: Spray inoculation with an aqueous suspension of zoospores. Selection of a proportion of the pods for chemical assay.

Day 3-5: Chemical assay of deposits on selected pods, together with comparable filter-paper samples sprayed at the same time as the pods.

Day 6: Counting of the numbers of lesions developed on the remainder of the pods, the pods having been stored since inoculation in a saturated atmosphere. The numbers of lesions on pods carrying varying amounts of fungicide are compared with the numbers on untreated pods. Mean values of the numbers of lesions for the various deposits are calculated and the significance of differences between means determined by a test. Plotting on log-probit paper of percentage infections for various fungicide deposits gives a dose response curve.

Day 7: Cleaning of apparatus. Possibly, chemical assay of deposits on pods on which lesions have been developed. (It has been found convenient for day 1 to be a Wednesday).

A number of commercially available fungicides have been investigated and the results obtained are in reasonable agreement with field experience. Further work will be delayed until the next wet season but, meanwhile, physical studies of fungicide formulations will continue, and a critical investigation of the laboratory sprayer will be carried out.

SOA.